SUPPLEMENTARY MATERIAL

Curcumin or bisdemethoxycurcumin for nose-to-brain treatment of Alzheimer disease? A bio/chemo-informatics case study

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Experimental

1. Construction of the virtual carrier using molecular dynamics simulations.

GROMACS (Pronk et al. 2013) v4.6.5 software package was used to carry-out all-atom molecular dynamics simulations . The atom typing and assignment of parameters and charges of the PLGA system according to CHARMM general force field (CgenFF) (Vanommeslaeghe et al. 2010) were carried-out online (https://cgenff.paramchem.org/). The simulated PLGA system contained 32 molecules of polylactic acid and 32 molecules of polyglycolic acid; each of them is composed of 35 monomers and is end capped with a methyl group. All systems were energy minimized using the steepest descent method. The PLGA system was then subjected to a molecular dynamics run, with full periodic boundary conditions, a time step of 2 fs, and a cut-off distance for Van der Waal's and electrostatic interactions of 1 nm. PME was used to calculate electrostatic interactions and LINCS algorithm was used to constrain all bonds. The system was equilibrated for 7 ns at 298K using a v-rescale thermostat at a pressure of 1 bar using a Berendsen barostat.

2. Obtaining the target peptides and proteins virtual matrices.

The crystal structure of the relevant nose-to-brain delivery and therapeutic targets were obtained from the protein data bank (http://www.rcsb.org). The following codes: 2ACM, 3G6I and 5IKQ corresponded for Mucin, P-gp efflux-pump receptor and cyclooxygenase inhibitor-2 (COX-2), respectively. The polar hydrogens were added to the obtained pdb files using MOE® version 2014.0901 (Chemical Computing Group Inc., Montreal, Canada).

3. Preparing the drugs chemical structures for docking.

The isomeric SMILES corresponding to the chemical structures of the studied drugs; curcumin and bisdemethoxycurcumin were obtained using PubChem®. The corresponding 3D chemical structures were generated using the builder function of MOE® version 2014.0901 (Chemical Computing Group Inc., Montreal, Canada). Further, energy minimization was carried out for all the investigated molecules using MMFF94x forcefield of the same software (Gooding et al. 2014;Costache et al. 2009).

4. Docking of the investigated drugs on the investigated carrier.

The docking analysis was employed using MOE version 2014.0901 (Chemical Computing Group Inc., Montreal, Canada). The pdb file of the protein nanoparticles matrix was imported to MOE where the identification of the binding site was performed using MOE's "Site finder" tool

(Elhefnawi et al. 2012) to be ready for docking using the "*triangle matcher"* as a placement method.

This software creates dummy atoms around the docking target atoms. These dummy atoms are considered the docking positions. ASE scores were utilized for calculating the binding energies scoring values. This score is proportional to the sum of the Gaussians $R_1R_2\exp(-0.5d^2)$ over all ligand atom - receptor atom pairs and ligand atom - alpha sphere pairs. ($R_{1)}$ and (R_2) are the radii of the atoms in Å, or is -1.85 for alpha spheres while (d) is the distance between the pair in Å. Because the ASE score makes good use of the previously stated Gaussian-type function to evaluate and optimize the overlap between the ligand and the site model, it can pose a ligand onto the docking site relatively faster and more effectively than using the other potential energy functions that are commonly used by other scoring algorithms. The posing stage through the use of the ASE score is followed by full atomistic energy minimization. Therefore, it is considered a very robust docking method (Goto et al. 2008). Like all commonly used scoring functions, lower binding energies (ΔG, kcal/mole) scores indicate more favourable interactions.

5. Calculating the main descriptors of the investigated drugs.

In order to explain the differences in docking scores observed for the studied drugs, some crucial constitutional, electronic and topological descriptors were calculated for the studied drugs. The selected descriptors were the molecular weight, LogP (O/W), total hydrophobic surface area, number of H-atoms donors and acceptors and finally the molecular flexibility. The descriptors were calculated using MOE version 2014.0901 (Chemical Computing Group Inc., Montreal, Canada) and utilizing the molecules SYBYL2 (mol2) files generated using ChemDraw® Ultra version 10.

6. Investigating the nose-to-brain Alzeheimer treatment potential of some proposed cucrumin analogs.

In order to extend our idea beyond the naturally occurring curcuminoids, newly proposed synthetic molecules (curcumin analogues) namely; diaminobisdemethoxycurcumin, dicarboxybisdemethoxycurcumin, diethoxybisdemethoxycurcumin, dihydroxybisdemethoxycurcumin and methylbisdemethoxycurcumin were constructed using ChemDraw Ultra v.10 (Cambridgesoft, Waltham, MA). Their corresponding SMILES are listed in Table 1. The corresponding Mol2 files needed for the subsequent docking on the investigated macromolecules and utilizing the software adopted in this study was obtained using Chem3D® Ultra version 10 (Cambridgesoft, Waltham, MA) after energy minimization using the MM2

force field of the same program. Docking of the constructed curcumin analogues were docked on the nano-carrier, proteins and peptides using MOE® version 2014.0901 (Chemical Computing Group Inc., Montreal, Canada). Moreover, some of the important physico-chemical, topological and constitutional descriptors of these molecules were computed using the same software.

7. Data Analysis of the obtained docking results using principal component analysis.

In order to correlate the docking results with the calculated physic-chemical, topological and constitutional descriptors of the investigated molecules, a multivariate statistical analysis technique *viz.* principal component analysis was utilized using JMP® software (SAS, Cary, NC). The results were demonstrated according to two principal components captured from and composed of the six investigated descriptors (Hathout 2014).

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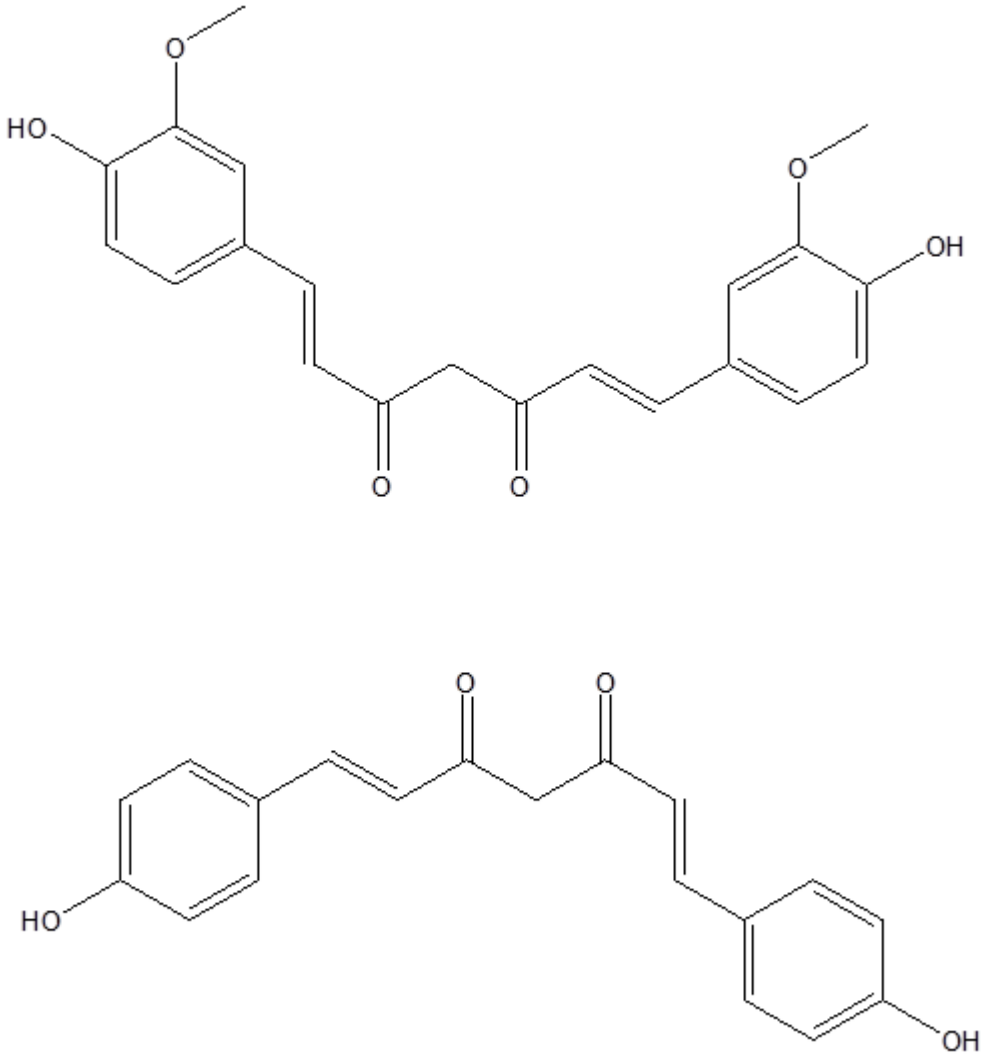
<u>Figure S1:</u> The chemical structure of (A) Curcumin and (B) Bisdemethoxycurcumin.



<u>Figure S2:</u> Successful docking of curcumin (left) and bisdemethoxycurcumin (right) on (A) PLGA polymer, (B) Mucin, (C) P-gp efflux pump, (D) Amyloid peptide and (E) Cyclooxygenase 2 enzyme.